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Chlamydial infection

Incidence in 'non-specific' urethritis

E. M. C. DUNLOP*†, J. D. VAUGHAN-JACKSON*†, S. DAROUGAR†‡ AND B. R. JONES†

From the Whitechapel Clinic, The London Hospital, and the Department of Clinical Ophthalmology† and the Virus Laboratory‡, Institute of Ophthalmology, London*

The isolation of *Chlamydia* in cell culture (Gordon, Harper, Quan, Treharne, Dwyer, and Garland, 1969) from urethral material from three of seven men who had presented because of acute non-specific urethritis (NSU) was reported by Dunlop, Hare, Darougar, Jones, and Rice (1969). Isolates were also obtained from cervical material from one of three sexual partners of these men. Isolation in yolk sac and the examination of smears were less sensitive tests for *Chlamydia* in clinical specimens than was culture in cell culture (Gordon and others, 1969; Darougar, Dwyer, Treharne, Harper, Garland, and Jones, 1971). Gordon and Quan (1971) obtained isolates from only nineteen of 84 men with NSU, but the urethral material had been obtained by different methods including a bacteriological loop which appeared to be relatively inefficient. Philip, Hill, Greaves, Gordon, Quan, Gerloff, and Thomas (1971) obtained isolates from seven of 31 men with non-gonococcal urethritis (NGU) using swabs and cotton-tipped applicators to collect urethral material. Ford and McCandlish (1971) isolated *Chlamydia* from urethral material obtained with a curette from only eight of 151 men suffering from NGU.

Using the simplified cell culture techniques of Darougar, Kinnison, and Jones (1971a, b), *Chlamydia* was grown from urethral material obtained by means of a curette, from eighteen (over 40 per cent.) of 41 men who had presented because of NSU and from cervical and other materials from five of 21 of the sexual partners of these men (Dunlop, Hare, Darougar, and Jones, 1971). This series has now been extended to comprise 99 men in whose cases valid tests for *Chlamydia* were made and 34 of their female sexual partners.

Patients and methods

108 heterosexual men who presented at the Whitechapel Clinic of The London Hospital, where they were found to be suffering from acute NSU, were tested, as were 34 of their sexual partners who were seen at the Institute of Ophthalmology. The techniques used for collecting material from the genital tract and from the rectum have been reported previously (Dunlop and others, 1964, 1965, 1969, 1971). The present procedure is outlined elsewhere in this issue (Dunlop, Vaughan-Jackson, and Darougar, 1972).

Only two patients of the 99 in whose cases valid tests for *Chlamydia* were made did not have any symptoms due to urethritis; one had a maximal cell count of 100 polymorphonuclear leucocytes per high-power (1/12″) field (HPF) in the urethral secretion after having refrained from micturition for 2 hours, the other a count of 100 in the overnight urethral secretion after 9½ hours.

All the remaining patients were suffering from acute non-specific urethritis. The maximal cell count in the urethral smear from each patient was 100 or more per HPF in 85 cases and less than 100 in fourteen; of this fourteen, in one case in which there was increased frequency of micturition the maximal count was only 20 per HPF 45 minutes after the patient had passed urine, in two cases the count was 30 after 2 hours, and in eleven it was 40 or more but less than 100.

One patient (Mr NS) was already using topical treatment from the Ophthalmic Department of another hospital for follicular conjunctivitis (Dunlop and others, 1971); examination showed punctate keratoconjunctivitis due to TRIC agent.

In addition, 34 female contacts of these men were tested for *Chlamydia*.

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Results

MEN WITH NSU

Chlamydia was isolated from urethral material from 44 of the 108 men. In the cases of nine patients both the cell culture inoculated with material from the endourethral swab and that inoculated with scrapings were reported as contaminated or otherwise unsatisfactory. Subtraction of these nine patients from the total leaves 99 patients who had valid tests (Table I, overleaf). *Chlamydia* was isolated from 44 (44·5 per cent.), all of whom had symptoms of urethritis.

TABLE I *Results of cell culture for* Chlamydia: *men suffering from NSU*

Valid tests: no. of patients	*Isolate positive: no. of patients*	*Isolate negative: no. of patients*
99	44 (44·5 per cent.)	55 (55·5 per cent.)

Appearances in the urethral meatus, resembling the follicles seen in the conjunctiva due to chlamydial infection, were seen with an operating microscope in only three cases, in all of which *Chlamydia* was isolated. Papillary congestion was marked in forty cases, in thirty of which *Chlamydia* was isolated. In two of the other cases in which *Chlamydia* was isolated the urethral meatus appeared entirely normal although the urethral secretion was purulent (150 polymorphonuclear leucocytes per HPF after the patient had refrained from micturition for 2 hours in one case and 100 per HPF after 2 hours in the other).

Only one of the men (Mr NS) who presented because of NSU had conjunctivitis. As already noted, he was receiving topical treatment ordered in the Ophthalmic Department of another hospital. Examination showed punctate keratoconjunctivitis due to TRIC agent. *Chlamydia* was isolated from his urethra and conjunctiva and from his wife's cervix and rectum.

Chlamydia was isolated from endourethral scrapings in 37 (39 per cent.) of 95 cases (Table II), from material obtained with an endourethral swab in 22 (37 per cent.) of sixty cases, and by a meatal swab in only 23 (26 per cent.) of 87 cases. The first two methods were of similar efficiency, the last was appreciably less efficient.

TABLE II *Results of different methods of collection of urethral material from men suffering from NSU*

Method	*No. of patients tested*	*Isolate obtained*	
		No.	*Per cent.*
Meatal swab	87	23	26
Endourethral swab	60	22	37
Endourethral scrape	95	37	39

There was weak fixation in a test for group complement-fixing antichlamydial antibody (LGVCFT) in sixteen cases, in thirteen of which *Chlamydia* was grown; but in the 31 other cases in which *Chlamydia* was isolated there was no fixation in the LGVCFT. When fixation was obtained, it was at a low level, the highest being complete fixation to only 1 in 8.

FEMALE CONTACTS

When the 34 female contacts of men suffering from NSU were tested, *Chlamydia* was isolated from ten (Table III), all of whom were sexual partners of men from whom that agent had been isolated.

TABLE III *Results of tests for* Chlamydia: 34 *female contacts of men suffering from NSU*

Site positive	*No. of patients with positive tests*
Any site	10
Cervix	10
Rectum	3 (of 33 tested)
Urethra	2 (of 11 tested)

The relationships of different findings in these 34 women to the isolation of *Chlamydia* are shown in Table IV (opposite).

Seven of them gave a history of vaginal discharge; *Chlamydia* was isolated from four of them. There was evidence of salpingitis in five, from two of whom *Chlamydia* was isolated. There were mucosal changes in the marginal area of the cervix in 21, from nine of whom *Chlamydia* was isolated from the cervix. 'Follicles' and collections of 'follicles' forming large lumps were seen in nine of the 21 women with mucosal changes; *Chlamydia* was isolated from five of them.

The cervical mucus was arbitrarily defined as purulent if it contained 25 or more polymorphonuclear leucocytes per HPF. It was purulent in 21 cases; *Chlamydia* was isolated from the cervix in six of these. In five cases the cervix appeared normal and smears did not contain pus; no isolate was obtained in these.

In one case there was papillary congestion of the urethral meatus; an isolate was obtained from this site. In three cases there were more than ten polymorphonuclear leucocytes per HPF in the urethral smear; *Chlamydia* was isolated from the urethra in one of these. The urethra appeared normal and the urethral smear contained less than ten polymorphonuclear leucocytes in seven cases; no isolate was obtained in these.

The anorectal mucosa was abnormal in two cases. In one there were many 'follicles' giving a 'cobblestone' appearance and in the other there was scarring and pus; *Chlamydia* was isolated from the rectum in both cases. Rectal mucus was arbitrarily defined as purulent if a smear contained ten or more polymorphonuclear leucocytes per HPF. By this defini-

TABLE IV *Findings in 34 female contacts of men suffering from NSU*

Finding	*No. of patients*	*No. of patients* Chlamydia-*positive*
History of vaginal discharge	7	4
Salpingitis	5	2
Cervix—Mucosal change	21	9
—Pus 25/HPF or more	21	6
—Normal, no pus	5	0
Urethra—Inflamed	1	1
—Pus 10/HPF or more	3	1
—Normal, no pus	7	0
Rectum—Mucosal change	2	2
—Pus 10/HPF or more	3	2
—Normal, no pus	29	0
LGVCFT fixation	11[a]	8
Associated infection	9[b]	2

[a]at low level, highest = complete fixation to 1 in 4
[b]candidiasis 6, trichomoniasis 2, previous syphilis 1

tion the mucus was purulent in three cases; isolates were obtained from anorectal material in two of these. The anorectal mucosa was normal and without pus in 29 cases; no isolate was obtained in these. Here, as at other sites, there was an association between the presence of signs of inflammation and the isolation of *Chlamydia*, and between the absence of signs of inflammation and the failure to isolate *Chlamydia*.

There was weak fixation in the LGVCFT in eleven cases; isolates were obtained in eight of these. Isolates were obtained in two cases in which there was no fixation in the LGVCFT.

Associated infection was present in nine cases: trichomoniasis in two, candidiasis in six, and previously treated syphilis in one.

Discussion

Culture of urethral material in irradiated McCoy cells has raised the proportion of cases in which *Chlamydia* can be demonstrated from 25 per cent. (Dunlop, Freedman, Garland, Harper, Jones, Race, du Toit, and Treharne, 1967) to over 40 per cent. There must be a large reservoir of chlamydial infection in the urethra in men and in the cervix, urethra, and rectum in women.

In one case of NSU (Mr NS), there was associated chlamydial infection of the eye that indicated that the isolates of *Chlamydia* obtained from the patient's urethra and from the cervix and the ano-rectal canal of his wife were isolates of TRIC agent.

Further studies of the 55 per cent. of cases of NSU in which *Chlamydia* cannot be demonstrated by cell culture will be of particular interest.

Summary

Valid tests of urethral material for *Chlamydia* were carried out in cell culture in the cases of 99 men who were suffering from NSU; *Chlamydia* was obtained from 44 (44·5 per cent.). Tests were carried out in 34 women who were sexual partners of these men; *Chlamydia* was isolated from ten who were all contacts of men from whom isolates had been obtained.

We are grateful to our colleagues who referred patients for investigation: to Dr. A. E. Wilkinson, Director of the V.D. Reference Laboratory, The London Hospital, for carrying out serological tests for syphilis, and cultures for bacteria, *Trichomonas vaginalis*, and *Candida*; to the late Prof. C. F. Barwell, and to other staff of the Virus Laboratory, The London Hospital, for carrying out the LGVCFT.

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Infection chlamydiale
Incidence dans l'urétrite non spécifique

SOMMAIRE

Des tests valables pour la recherche des *Chlamydia* en culture de cellules furent pratiqués avec le matériel urétral de 99 hommes atteints d'urétrite non spécifique et des *Chlamydia* furent obtenus pour 44 d'entre eux (44,5 pour cent). Les tests furent pratiqués chez 34 femmes, partenaires sexuelles de ces hommes; les *Chlamydia* furent isolés pour 10 d'entre elles, toutes partenaires d'hommes chez lesquels l'agent avait été isolé.